

# Elevated mir-141 in obesity: Insights into the interplay with sirtuin 1 and non-alcoholic fatty liver disease

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## Abstract

**Background:** Changes in gene expression related to obesity are linked to microRNAs, such as miR-141, which play a crucial role in metabolic homeostasis. Sirtuin 1 (SIRT1), an enzyme that plays a crucial role in regulating various cellular functions and metabolism, is implicated in obesity and the ensuing non-alcoholic fatty liver disease (NAFLD). The aim of this research was to evaluate the levels of miR-141 and its relationship with SIRT1 and NAFLD.

**Methods:** A group of 100 adults (50 with obesity and 50 with normal-weight) were selected and underwent complete clinical evaluation and anthropometric measurements. Biochemical parameters were assessed in blood serum, and the levels of miR-141 in plasma were measured by real-time PCR. The expression of the SIRT1 gene was also evaluated in the peripheral blood mononuclear cells using Real-time PCR. The ELISA technique was used to determine insulin levels. Liver steatosis was assessed by ultrasound.

**Results:** The results showed that levels of miR-141 were significantly increased in participants with obesity compared with the control group. Conversely, the expression of the SIRT1 gene in individuals with obesity was lower than that in control participants. A strong negative correlation was observed between miR-141 and SIRT1 and a strong positive association was observed between miR-141 and metabolic parameters. Furthermore, participants with fatty liver had significantly elevated levels of miR-141 gene expression and lower expression of SIRT1 gene, compared to those without fatty liver.

**Conclusion:** elevated levels of miR-141 in individuals with obesity might be a contributing factor in the repression of SIRT1 in obesity and its consequences, including NAFLD. Therefore, miR-141 might serve as a suitable diagnostic and therapeutic target in obesity and NAFLD.

## KEYWORDS

microRNA, miR-141, non-alcoholic fatty liver disease, obesity, sirtuin1

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## 1 | INTRODUCTION

The increasing prevalence of obesity has had a dramatic impact on the occurrence of obesity-related illnesses, one of which is non-alcoholic fatty liver disease (NAFLD), first described in 1980.<sup>1-3</sup> NAFLD is characterized by the excessive accumulation of lipids in liver cells, disrupting metabolic processes and leading to inflammation, insulin resistance, oxidative stress, liver fibrosis, and even hepatocellular carcinoma.<sup>4,5</sup> Although various metabolic factors involved in the progression of NAFLD have been identified, the underlying mechanisms are not fully understood. Recent research has linked certain microRNAs to obesity and its associated metabolic disturbances.<sup>6</sup>

MicroRNAs (miRNAs) are naturally occurring, small non-coding RNA molecules approximately 21–25 nucleotides in length. MicroRNAs regulate gene expression by binding to the 3'-untranslated region (3'UTR) of mRNA molecules, thereby repressing translation and potentially causing mRNA cleavage and deacetylation.<sup>7,8</sup> miRNAs play a crucial role in lipid metabolism, particularly fatty acids and cholesterol, glucose homeostasis, insulin signaling, and the expression of key regulatory enzymes.<sup>8-11</sup> Furthermore, miRNAs are involved in both normal biological processes, such as organismal development, metabolism, and physiology, and the pathogenesis of diseases such as diabetes, cancer, cardiovascular disease, and inflammation.<sup>12</sup> Several miRNAs have been implicated in liver pathophysiology, including NAFLD, cirrhosis, and hepatocellular carcinoma.<sup>13</sup> Among these, miR-141, a member of the miR-200 family, has emerged as an miRNA with potential roles in liver physiology and pathology.<sup>14-16</sup> miR-141 has been proposed as a biomarker for various liver disorders and as a marker for liver aging.<sup>17</sup>

Sirtuins comprise a group of enzymes that play an important regulatory role in various cellular processes, especially carbohydrate and lipid metabolism. Sirtuin1 (SIRT1), an NAD-dependent deacetylase enzyme, has been shown in recent years to be dysregulated in obesity and seems to play an important role in the pathophysiology of NAFLD.<sup>18,19</sup> Various mechanisms relate SIRT1 function to protection against NAFLD. Autophagy, which is an important parameter in many disease states including NAFLD, is modulated by SIRT1. For instance, SIRT1 augmentation by active components of ginseng root can ameliorate NAFLD by restoring autophagic flux. Additionally, SIRT1 affects metabolism in liver cells by increasing CPT1A level and decreasing triglyceride levels in HepG2 cells. SIRT1 can also attenuate oxidative stress, suppress mitochondrial fission, improve mitochondrial biogenesis, and decrease hepatocyte oxidative stress by deacetylating Nrf2.<sup>20</sup>

Generally, the expression of SIRT1 in liver cells is high, and its inhibition leads to increased glucose and lipid levels.<sup>21</sup> Low levels of SIRT1 have been reported in animal models of obesity and patients with obesity, especially those with insulin resistance, diabetes, and NAFLD.<sup>19,22</sup>

According to a bioinformatics study, miR-141 aligns with the 3'UTR region of sirtuin1 (SIRT1). Our previous experimental study

confirmed this binding and revealed that upregulation of miR-141 could suppress SIRT1 expression and induce lipid accumulation in cultured hepatocytes, and that it can directly target and align with the 3'-UTR of SIRT1 mRNA.<sup>23</sup> The aim of this study was to investigate the levels of miR-141 together with SIRT1 gene expression in participants with obesity and their relationship with insulin resistance, liver function, and metabolic syndrome (MetS) parameters with the hypothesis that miR-141 might be responsible for the suppression of SIRT1 expression in participants with obesity and subsequent NAFLD.

## 2 | MATERIAL AND METHODS

### 2.1 | Participants

This study involved 100 participants, an obesity group composed of with 50 participants with obesity (BMI over 30), while the remaining half consisted of healthy individuals with a normal weight who served as the control group. Prior to enrollment, written informed consent was obtained from each individual. Patients were carefully selected from those visiting the clinics of the Institute of Endocrinology and Metabolism or the Minimally Invasive Surgery Research Center, while healthy controls were chosen from volunteers following clinical evaluation. The control subjects did not have extensive recent weight loss, and none of them had undergone any process for weight loss, such as bariatric surgery. Individuals with various chronic and acute medical conditions, such as heart and kidney diseases, infections, malignancies, and inflammatory conditions, as well as those taking medications that could affect lipid and carbohydrate metabolism, were excluded from the study. Specific investigations were conducted to rule out abnormalities in adrenal and thyroid hormones, known endocrine causes of obesity, as well as diabetes. The participants were not following any special diet and had normal levels of physical activity and muscle mass. Pregnant and lactating individuals were not included. All participants underwent comprehensive examinations by a single clinician, which included documenting their height, weight, waist and hip measurements, blood pressure, and family medical history. Waist circumference was measured by a single clinician, using a non-elastic measuring tape, with the patient in the standing position, at the midpoint between the lower margin of the last palpable rib and the top of the iliac crest.

To assess MetS based on IDF standards,<sup>24</sup> specific factors were taken into consideration, including abdominal obesity (waist circumference greater than 102 cm for males and 88 cm for females), serum triglyceride levels exceeding 150 mg/dl, HDL cholesterol levels below 40 mg/dl, systolic hypertension (over 130 mmHg), diastolic blood pressure above 85 mmHg, and fasting plasma glucose levels above 110 mg/dl. Individuals who met three or more of these characteristics were diagnosed with MetS. Ultrasonography was performed on the liver to evaluate the degree of hepatic steatosis.

## 2.2 | Sample collection and preparation

Blood samples were collected from each individual following an overnight fast and immediately divided into two tubes. One tube contained EDTA (ethylenediaminetetraacetic acid) as an anticoagulant for the separation of peripheral blood mononuclear cells (PBMCs), while the other tube was plain for the separation of serum. After centrifugation, the separated serum samples were frozen and stored at  $-80^{\circ}\text{C}$  for future analyses.

To isolate mononuclear cells from both patients and healthy donors, fresh samples were processed using a Ficoll-Hypaque density gradient (Sigma-Aldrich, USA). The isolated cells were stored in tubes that were free from RNase and DNase contamination. Briefly, 10 mL of peripheral blood samples diluted with phosphate buffer saline (PBS) were layered over 3 mL of Ficoll-Hypaque. The tubes were then centrifuged for 20 min at 5000 RPM, allowing for the isolation of PBMCs from the interlayer. The PBMCs were subsequently washed twice with PBS.

## 2.3 | Biochemical measurements

Biochemical parameters, including fasting blood glucose (FBS), triglyceride (TG), total cholesterol (TC), high-density lipoprotein cholesterol (HDL-C), low-density lipoprotein cholesterol, aspartate aminotransferase (AST), alanine aminotransferase (ALT), alkaline phosphatase, total bilirubin, and direct bilirubin, were measured using commercially available enzymatic colorimetric kits (Pars Azmoon, Iran) and analyzed on a Hitachi autoanalyzer. Insulin levels were measured using the enzyme-linked immunosorbent assay (ELISA) Kit (Monobind, USA).

## 2.4 | Gene expression assessments

Separate RNA purification was conducted from both plasma and PBMCs. To extract total RNA from plasma, which contains small RNA molecules of less than 100 nucleotides, a sample volume of 200  $\mu\text{L}$  was processed using the GeneAll Hybrid-R Blood RNA isolation kit

(GeneAll, Korea). For RNA purification from PBMCs, the Hybrid-R miRNA isolation kit (GeneAll, Korea) was utilized. This kit is optimized for the extraction of small RNA molecules ( $<200$  nucleotides) following cell lysis with RiboEx lysis reagent. To initiate cDNA synthesis from miRNAs, a poly(A) tail was added to elongate the miRNAs using Poly(A) Polymerase (New England Biolabs). The High-Capacity cDNA Reverse Transcription Kit (Applied Biosystems, USA) was used for cDNA synthesis, employing a primer containing a complementary sequence for the poly(A) tail and an adapter sequence (refer to Table 1 for details).

Real-time quantitative PCR was utilized to investigate the levels of miR-141 and SIRT1 mRNA in plasma and PBMCs, respectively, using SYBR Premix EX Taq II (Takara, Japan). The reference genes used in this assay were miR-16 and GAPDH, which served as normalization controls. A reverse primer, complementary to the adapter sequence, was employed for amplifying miR-141. The primer sequences can be found in Table 1. Cel-miR-39-1 was added to the samples as a Spike-In control, using its primer for amplification, to ensure reproducibility. A standard curve containing  $1 \times 10^3 - 1 \times 10^6$  copies of Cel-miR-39 cDNA was generated independent of a serum/plasma sample and RNA purification procedure, allowing estimation of the recovery of miRNeasy Serum/Plasma Spike-In Control.

Gene expression quantification was performed using the  $\Delta\text{Ct}$  method, which involved subtracting the Ct value of the reference gene from the Ct value of the target gene.

## 2.5 | Statistical analysis

The normal distribution of the data was assessed using the Kolmogorov-Smirnov test. Variables that exhibited a normal distribution were analyzed using the independent sample *t*-test and analysis of variance, followed by Tukey's post hoc test. For variables that did not follow a normal distribution, the Mann-Whitney *U* test or Kruskal-Wallis test was applied. The association between variables was examined using either Pearson or Spearman correlation analysis. Statistical analysis was conducted using IBM SPSS version 24 and MedCalc version 18.9.10 (Ostend, Belgium). A significance level of  $p < 0.05$  was considered to indicate a statistically significant difference.

TABLE 1 The sequence of primers.

Primers	Sequence
miR-141-3p	5'-CGTAACACTGTCTGGTAAAGATGG -3'
miR-16-5p	5'-GGGTAGCAGCACGTAATATTGG -3'
Adapter	5'-CGAGCACAGAATTAATACGACTCACTATAGGTTTTTTTTTTTAG -3'
Universal	5'-GCGAGCACAGAATTAATACGACTC -3'
SIRT1-F	5'-TGCGGGAATCAAAGGATAA -3'
SIRT1-R	5'-CAGGCAAGATGCTGTTGCA -3'
GAPDH-F	5'-GGGAAGGTGAAGGTCGGAGT-3'
GAPDH-R	5'-TCCACTTTACCAGAGTAAAAGCAG -3'

## 2.6 | Institutional board approval

This study was approved by the Ethics Committee of Iran University of Medical Sciences (Number 94-05-30-27493).

## 3 | RESULTS

The anthropometric characteristics of the study groups are shown in Table 2. The participants were matched in terms of gender, age, and height, and there were no significant differences between the two groups. Individuals with obesity exhibited significant increases in various anthropometric indices, including weight, BMI, waist and hip

TABLE 2 Anthropometric characteristics of the studied subjects.

Variable	Obese	Control	p-value
Gender (M/F)	26/24	16/34	0.043
Age (Year)	43.1 ± 17.7	40.4 ± 10.7	0.22
Height (cm)	168.4 ± 8.6	172.1 ± 7.4	0.025
Weight (kg)	105.4 ± 21.7	71.6 ± 10.7	<0.001
BMI	36.6 ± 7.3	23.7 ± 3.2	<0.001
WC (cm)	111.5 ± 17.4	85.8 ± 8.9	<0.001
HC (cm)	123.9 ± 19.9	96.2 ± 6.9	<0.001
WHR	0.9 ± 0.03	0.89 ± 0.05	0.28
SBP (mmHg)	122.4 ± 5.9	119.4 ± 2.3	<0.001
DBP (mmHg)	82.8 ± 6.8	78.8 ± 3.2	<0.001
FBS (mg/dl)	101.4 ± 25.6	91.9 ± 17.8	0.031
TG (mg/dl)	146.1 ± 82.6	153.8 ± 103.9	0.68
TC (mg/dl)	175.4 ± 33.8	178.06 ± 32.3	0.69
LDL-C (mg/dl)	98.08 ± 20.08	102.8 ± 18.9	0.31
HDL-C (mg/dl)	41.1 ± 10.04	40.08 ± 7.9	0.85
Insulin (mIU/L)	11.3 ± 8.2	8.8 ± 6.3	0.098
HOMA-IR	3.09 ± 3.1	2.08 ± 1.6	0.044
AST (IU/L)	31.5 ± 21.3	26.02 ± 8.7	0.095
ALT (IU/L)	31.08 ± 25.5	30.7 ± 17.9	0.44
ALP (IU/L)	206.5 ± 47.6	193.2 ± 28.6	0.095
T bilirubin (mg/dl)	0.71 ± 0.47	0.73 ± 0.35	0.83
D bilirubin (mg/dl)	0.24 ± 0.12	0.24 ± 0.08	0.92

Note: Values are expressed as mean ± SD.

Abbreviations: ALP, alkaline phosphatase; ALT, alanine aminotransferase; AST, aspartate aminotransferase; BMI, body mass index; D bilirubin, direct bilirubin; DBP, diastolic blood pressure; FBS, fasting blood glucose; HC, hip circumference; HDL-C, high density lipoprotein cholesterol; HOMA-IR, homeostatic model assessment of insulin resistance; LDL-C, low-density lipoprotein cholesterol; SBP, systolic blood pressure; T bilirubin, total bilirubin; TC, total cholesterol; TG, triglycerides; WC, waist circumference; WHR, waist-to-hip ratio.

circumference, as well as systolic and diastolic blood pressures. Among the biochemical factors, FBS and HOMA-IR were significantly higher in the obesity group compared to the control participants, while other biochemical parameters did not differ significantly between the two groups (Table 2).

In the obesity group, 21 (49%) patients were diagnosed with MetS, and 38 (76%) patients showed varying degrees of liver lipid accumulation. Among them, 36% had Grade 1, 46% had Grade 2%, and 18% had Grade 3 fatty liver. Table 3 presents a comparison of anthropometric indices and biochemical parameters in the studied participants based on the degree of hepatic steatosis.

The expression of miR-141 in plasma was compared between the two groups, and the results revealed that individuals with obesity exhibited increased levels of miR-141 compared to normal-weight individuals ( $p < 0.001$ ) (Figure 1A). miR-141 was also compared in participants diagnosed with and without MetS, and the results showed that miR-141 was significantly increased in MetS ( $p < 0.05$ ) (Figure 1B). The same comparison was performed for SIRT1, and its expression was found to be significantly decreased in participants with obesity ( $p < 0.001$ ) (Figure 1c). However, there was only a slight decrease in SIRT1 expression among individuals with MetS, which did not yield a statistically significant difference ( $p = 0.3$ ) (Figure 1D).

The levels of circulatory miR-141 were compared in the studied groups according to the degree of hepatic steatosis. miR-141 levels showed an increasing trend with increased liver steatosis, and the expression of miR-141 was significantly increased in patients with Grade 2 and 3 NAFLD (Figure 1E). On the contrary, the results of the expression of the SIRT1 gene displayed a significant decline along with increased lipid accumulation in the liver, and significantly reduced expression of SIRT1 was observed in the various grades of NAFLD compared to the control group (Figure 1F).

In the correlation analysis, a positive association was observed between miR-141 and all parameters of metabolic dysregulation, including BMI, WC, HC, SBP, DBP, FBS, TC, HDL-C, insulin, HOMA-IR, and AST (Table 4). On the other hand, the relationship between SIRT1 and obesity parameters, glycemic indices, lipid profile, and liver function tests, including ALT and AST, was negative. When correlation analysis was performed only in the obesity group, the relationship between SIRT1 and miR-141 with most parameters still remained significant, except with SBP, DBP, and FBS for SIRT1, and AST for miR-141. The correlation analysis also revealed a significant reverse correlation between SIRT1 and miR-141 ( $r = -0.801$  in all participants and  $r = -0.813$  in those with obesity), which remained significant after controlling for BMI ( $p < 0.001$ ) (Figure 2).

## 4 | DISCUSSION

This study found that overweight individuals had elevated levels of miR-141 compared with those of average body weight. A significant correlation between miR-141 and BMI and WC was also found. Although miR-141 has not been previously investigated in patients with obesity, there is evidence showing that many other microRNAs

**TABLE 3** Comparison of anthropometric indices between groups with different degrees of hepatic steatosis.

Variable	Normal	Grade 1	Grade 2	Grade 3
Number	61	14	17	7
Age	40.68 ± 10.92	45.5 ± 15.3	43.72 ± 10.51	38.71 ± 6.99
BMI	25.13 ± 4.6	37.14 ± 6.97***	38.58 ± 7.6***	39.12 ± 8.45***
WC (cm)	88.31 ± 10.8	110.29 ± 13.37***	118.28 ± 18.94***	115.29 ± 21.3***
HC (cm)	99.02 ± 9.92	123.29 ± 17.5***	130.44 ± 23.14***	127.57 ± 20.67***
WHR	0.89 ± 0.05	0.89 ± 0.04	0.9 ± 0.04	0.89 ± 0.02
SBP (mmHg)	119.25 ± 2.18	122.86 ± 8.25	123.33 ± 5.94*	122.83 ± 4.88
DBP (mmHg)	79.05 ± 3	82.86 ± 8.25	83.33 ± 8.4*	82.86 ± 4.88
FBS (mg/dl)	91.6 ± 16.4	99.4 ± 18.2	104.05 ± 21.8	116.4 ± 49.4*
TG (mg/dl)	154.3 ± 97.4	149.8 ± 121.3	147.3 ± 71.3	119 ± 41.3
TC (mg/dl)	173.8 ± 32.03	158.7 ± 26.9	189.7 ± 32.3	204.24 ± 29.4
LDL-C (mg/dl)	99.6 ± 19	89.2 ± 18.7	107.1 ± 18.6	116.8 ± 14.7
HDL-C (mg/dl)	39.7 ± 7.5	36.5 ± 7.3	44.8 ± 10.5	51.2 ± 10.02*
Insulin (mIU/L)	8.6 ± 6	10.44 ± 6.31	12.8 ± 10.2	15.26 ± 9.13
HOMA-IR	2.01 ± 1.52	2.72 ± 2.14	3.66 ± 4.13	4.58 ± 3.32*
AST (IU/L)	25.72 ± 8.9	22.71 ± 5.59	27.88 ± 11	69.71 ± 33.46***
ALT (IU/L)	29 ± 17.14	24 ± 10.92	33.22 ± 17.94	79.14 ± 32.95***
ALP (IU/L)	189.33 ± 33.8	212.85 ± 53.36	212.38 ± 40.58	231.52 ± 24.42*
T bilirubin (mg/dl)	0.71 ± 0.34	0.67 ± 0.31	0.73 ± 0.59	0.88 ± 0.65
D bilirubin (mg/dl)	0.23 ± 0.08	0.23 ± 0.04	0.23 ± 0.08	0.35 ± 0.26*

Note: Values are expressed as mean ± SD.

Abbreviations: ALP, alkaline phosphatase; ALT, alanine aminotransferase; AST, aspartate aminotransferase; BMI, body mass index; D bilirubin, direct bilirubin; DBP, diastolic blood pressure; FBS, fasting blood glucose; HC, hip circumference; HDL-C, high density lipoprotein cholesterol; HOMA-IR, homeostatic model assessment of insulin resistance; LDL-C, low-density lipoprotein cholesterol; SBP, systolic blood pressure; T bilirubin, total bilirubin; TC, total cholesterol; TG, triglycerides; WC, waist circumference; WHR, waist-to-hip ratio.

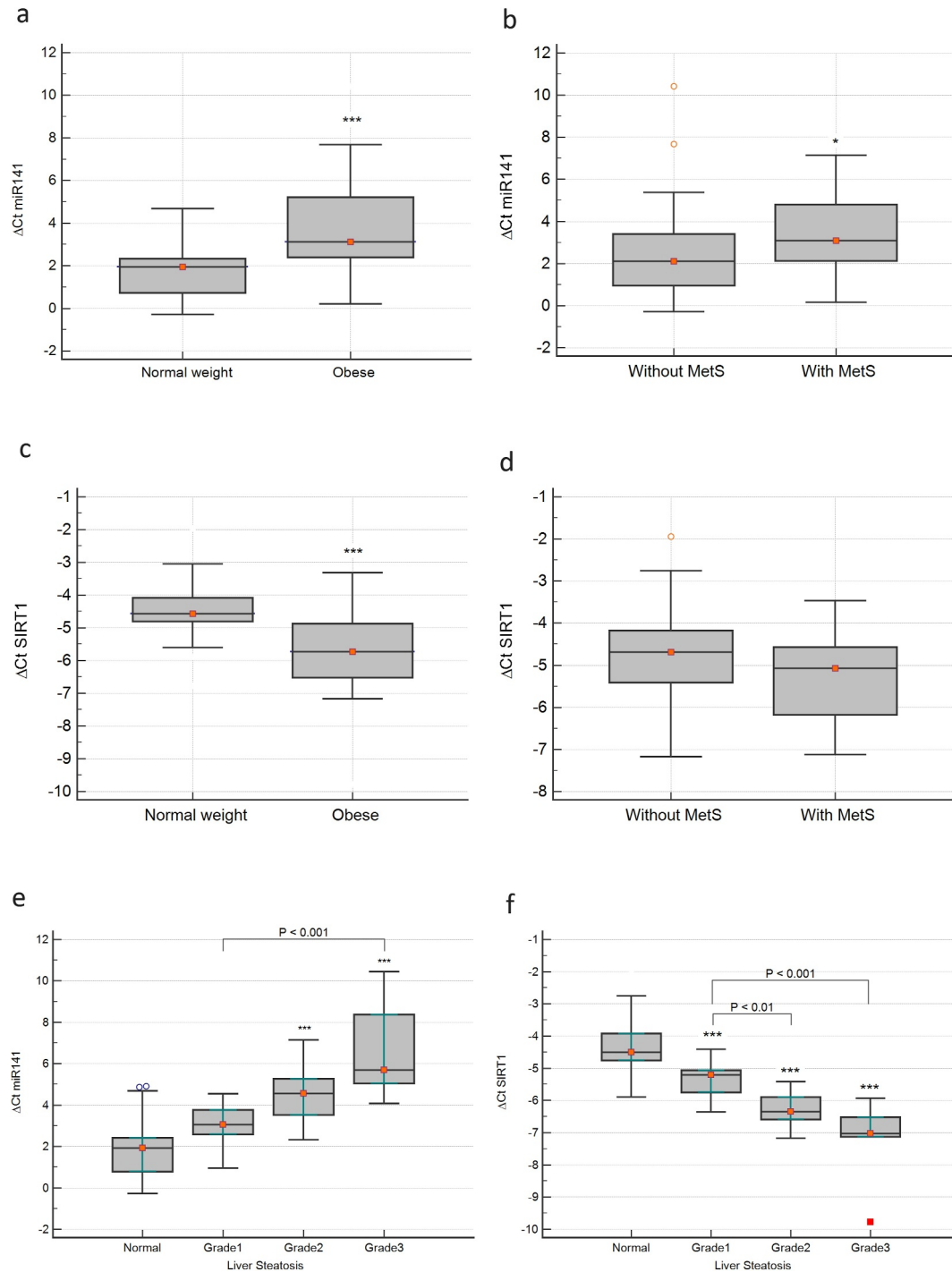
\*\*\* $p < 0.001$ , \*\* $p < 0.01$ , \* $p < 0.1$ .

are disrupted in obesity. These alterations have profound effects on the processes related to obesity, such as adipogenesis, lipid metabolism, and inflammation.<sup>25</sup> For example, miR-141-3p has been found to be significantly up-regulated in mice fed a high-fat diet, and it has been shown to modulate ATP production and induce oxidative stress.<sup>26</sup> In addition, miR-141 has been found to be increased in pre-eclampsia and is involved in inflammation associated with this disorder.<sup>27</sup>

MicroRNAs have also been introduced to be responsible for various disorders associated with obesity, including endothelial dysfunction, cardiovascular diseases, insulin resistance, diabetes, MetS, and NAFLD.<sup>25</sup> In our study, we evaluated MetS and NAFLD in the participants and found that miR-141 was significantly increased in both conditions. Furthermore, the level of miR-141 showed a consistent increase with the severity of hepatic steatosis. We also observed significant correlations between miR-141 and MetS components, such as insulin resistance indices and blood pressure. Consistently, Li et al. identified 40 upregulated and 10 downregulated miRNAs in patients with MetS.<sup>28</sup> In line with our results, miR-141 has been associated with

insulin resistance, a key component of MetS. Elevated levels of miR-141 have been observed in diabetic mice and elderly individuals with diabetes, and treatment with pioglitazone, a widely used therapy for insulin resistance, has been shown to correct miR-141 expression. The increased expression of miR-141 has been linked to impaired insulin secretion and reduced proliferation of pancreatic  $\beta$  cells. Conversely, suppressing miR-141 in  $\beta$  cells has been found to decrease the damage associated with type 2 diabetes.<sup>29</sup>

In the case of fatty liver, the expression of certain microRNAs is reduced (e.g., miR-22), while others are increased (e.g., miR-34a and miR-146b). These changes in expression levels ultimately affect the expression of genes involved in liver fat metabolism, contributing to the development of fatty liver.<sup>30</sup> In hepatocytes, miR-141 has been extensively studied and its inhibition has been shown to suppress HSC activation via the AKT/mTOR pathway by targeting PTEN. This identified miR-141 as a potential therapeutic target for the treatment of liver fibrosis.<sup>31</sup> In our previous research, we had demonstrated that inhibiting miR-141 could reduce lipid accumulation in hepatocytes, while its upregulation resulted in increased intracellular



**FIGURE 1** Comparison of miR-141 between the obesity and control groups (A), and the participants with or without metabolic syndrome (MetS) (B). Comparison of *SIRT1* gene expression between the obesity and control groups (C), and the participants with or without MetS (D). The comparison of miR-141 (E) and *SIRT1* (F) gene expression of the different groups categorized based on the grade of liver steatosis. \* $p < 0.05$ , \*\* $p < 0.01$ , \*\*\* $p < 0.001$ .

lipid accumulation.<sup>23</sup> Tran et al. also showed that suppressing miR-141 levels led to reduced hepatic steatosis and inflammation by reprogramming lipid and inflammation signaling pathways.<sup>32</sup>

*SIRT1* is the most extensively studied sirtuin and is implicated in both alcoholic and non-alcoholic fatty liver diseases. Increasing *SIRT1* activity can decrease fat accumulation in adipocytes, enhance mitochondrial energy production, and promote fatty acid oxidation.<sup>33</sup>

Additionally, *SIRT1* plays a beneficial role in regulating hepatic lipid metabolism, managing hepatic oxidative stress, and modulating hepatic inflammation. In patients with obesity presenting with severe fatty liver, *SIRT1* levels are reduced in visceral fats, which can have a negative impact on mitochondrial function.<sup>34</sup> It has been demonstrated that *SIRT1* is downregulated in children and adolescents with obesity,<sup>22</sup> although the exact mechanism of its suppression is not



**TABLE 4** Correlation coefficients indicating the relationship of Sirt1 and miR-141 with biochemical indices.

Variable	All subjects		Participants with obesity	
	SIRT1	miR-141	SIRT1	miR-141
Age	-0.009	0.099	0.027	0.133
BMI	-0.509**	0.533**	-0.314*	0.445**
WC (cm)	-0.489**	0.247**	-0.405**	0.438**
HC (cm)	-0.468**	0.489**	-0.294*	0.406*
WHR	-0.139	-0.058	-0.178	0.411*
SBP (mmHg)	-0.238*	0.31**	-0.235	0.289
DBP (mmHg)	-0.214*	0.294*	-0.117	0.289
FBS (mg/dl)	-0.333**	0.25*	-0.222	0.358*
TG (mg/dl)	-0.024	0.080	0.170	-0.055
TC (mg/dl)	-0.264**	0.246*	-0.517**	0.346*
LDL-C (mg/dl)	-0.209*	0.191	-0.604**	0.359*
HDL-C (mg/dl)	-0.293**	0.289*	-0.558**	0.374*
Insulin (mIU/L)	-0.324**	0.285*	-0.336*	0.369*
HOMA-IR	-0.346**	0.338**	-0.354*	0.432**
AST (IU/L)	-0.317**	0.358**	-0.402**	0.278
ALT (IU/L)	-0.274**	0.215	-0.461**	0.195
ALP (IU/L)	-0.174	0.2	-0.242	0.302
T bilirubin (mg/dl)	-0.034	-0.013	-0.122	0.031
D bilirubin (mg/dl)	-0.149	0.15	-0.238	0.226

Note: Values are expressed as mean  $\pm$  SD.

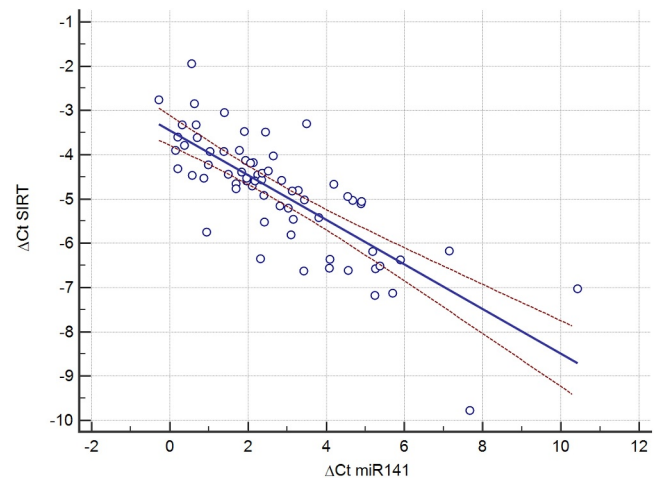
Abbreviations: ALP, alkaline phosphatase; ALT, alanine aminotransferase; AST, aspartate aminotransferase; BMI, body mass index; D bilirubin, direct bilirubin; DBP, diastolic blood pressure; FBS, fasting blood glucose; HC, hip circumference; HDL-C, high density lipoprotein cholesterol; HOMA-IR, homeostatic model assessment of insulin resistance; LDL-C, low-density lipoprotein cholesterol; SBP, systolic blood pressure; T bilirubin, total bilirubin; TC, total cholesterol; TG, triglycerides; WC, waist circumference; WHR, waist-to-hip ratio.

\*\*\* $p < 0.001$ , \*\* $p < 0.01$ , \* $p < 0.1$ .

fully understood. Although we had limitations in extending the sample size, low levels of SIRT1 were observed in participants with obesity, which correlated well with the levels of miR-141. Therefore, the decreased levels of SIRT1 in obesity may be attributed to the increased levels of miR-141. This notion is supported by a previous in vitro study showing that miR-141 repressed SIRT1 activity and expression, as well as a bioinformatics exploration indicating alignment of miR-141 with the 3'-UTR of SIRT1 mRNA.<sup>23</sup>

## 5 | CONCLUSION

The results of the current study demonstrated elevated levels of miR-141 in individuals with obesity, which were associated with MetS and hepatic steatosis. There was a correlation between miR-



**FIGURE 2** The correlation between the expression of SIRT1 gene and miR-141.

141 and SIRT1 expression, as well as metabolic parameters. Given the significant role of SIRT1 in obesity and its related conditions, such as NAFLD, miR-141 could be regarded as a potential target for addressing disorders associated with obesity; however, further experiments are required to establish the translational value of miR-141 in the context of obesity.

## ACKNOWLEDGMENTS

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## CONFLICT OF INTEREST STATEMENT

The authors declare no conflicts of interest.

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